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Page 8 second paragraph

A DNA-sequence encoding for the novel enzyme as defined in the patent claims is another aspect of the present invention. Preferably the DNA-sequence has a nucleotide sequence as given in the [SEQ ID NO:1 or 2] SEQ ID NO:1 or 3. The DNA sequence according to the present invention is preferably including a promoter and contained in an expression vector such as plasmid, a cosmid or a virus.

Paragraph bridging pages 19-20

cDNA was made from 250 ng of the isolated poly A^+ RNA using SMART™ PCR cDNA Library Construction kit (Clontech) according to the protocol recommended by the manufacturer. In brief, 1st strand cDNA was made by combining 250 ng A+ RNA with 10 pmol SMART oligonucleotide (5'-TACGGCTGCGA GAAGACGACAGAAGGG-3') (SEQ ID NO:5) and 10 pmol CDS/3' PCR primer (Oligo(dT)30 N-1N (N=A, G, C, or T; N-1 = A, G, C or T)) in a final volume of 5 µl, and incubated at 72°C for 2 min and then placed directly on ice for 2 min to denature the RNA. Then enzyme and buffer were added to the reaction mixture to a final volume of 10 µl, consisting of 50 mM Tris/HCl. pH 8.3, 6mM $MgCl_2$, 75mM Kcl, 2mM DTT, 1mM dATP, dCTP, dGTP and dTTP respectively and 200 U SuperScript™ II reverse transcriptase (Gibco BRL), and then incubated at 42°C for 1h. Synthesis of 2nd strand was done by PCR in a final volume of 100 µl, containing 2 µl of the 1st strand reaction as template, 40 mM Tricine/KOH pH 9.2 (25°C), 15 mM KOAc, 3.5 mM $Mg(OAc)_2$, 3.75 µg/ml BSA, 0.2mM of dATP, dCTP, dGTP and dTTP respectively, 1U Advantage cDNA Polymerase Mix (Clontech), 0.2 µM 5'-PCR primer (5'-TACGGCTCCGAGAAGACGACAGAA-3') (SEQ ID NO:6) and CDS/3'-PCR primer respectively.

Paragraph bridging pages 20 and 21

Degenerated oligonucleotide primers were designed from two conserved regions (GQDPYH and VFLLWG) from known UNG-amino acid sequences. Codon usage for Atlantic cod were also considered when designing the primers. The UNG fragment was generated by PCR with cod liver cDNA as template in a final volume of 50 μl, containing 10 mM Tris/HCl pH 9.0 (25°C), 50 mM KCl, 0.1% Triton X-100, 10 ng cDNA, 0.2 mM dATP, dCTP, dGTP and dTTP respectively, 2.0 μM upstream primer (5'-GGH-CAR-GAY-CCC-TAY-CA-3') (SEQ ID NO:7) and downstream primer (5'-DCC-CCA-SAG-SAG-RAA-VAC-3')[1] (SEQ ID NO:8) respectively and 2.5 U Taq-polymerase (Promega). PCR was carried out [by] at 94°C for 1 min, 60°C for 1 min and 72°C for 1 min, and a final extension step of 72°C for 5 min.

Paragraph bridging pages 21 and 22

The sequence deduced from the 300 bp fragment of the UNG-gene was used to design two primers for both 3'- and 5'- rapid amplification of cDNA ends (RACE), with a small overlap region between the two fragments generated. Both 3'- and 5'- RACE reactions were done in a volume of 50 μl with 1 μl of diluted cDNA with RACE-adaptors as template, 0.2 μM internal 3'-(5'-TGTACCGACATTGATGGCTTCAAGCAT-3') (SEQ ID NO:9) or 5'-(5'-CCCATCCGCTTAGATCTCCATGTCCAG-3') (SEQ ID NO:10) RACE primers, respectively, 0.2 μM AP1-primer (supplied by manufacturer) (5'-CCATCCTAATACGACTCACTATAGGGC-3') (SEQ ID NO:11), 40 μM Tricine/KOH pH 9.2 (25°C), 15 mM KOAc, 3.5 mM Mg $(OAc)_2$, 3.75 μg/ml BSA, 0.2 mM of each dATP, dCTP, dGTP and dTTP and 10U Advantage cDNA Polymerase Mix (Clontech). Amplification was done in a GeneAmp 9700 thermocycler (Perkin Elmer), 94°C for 30 sec followed by 5 cycles of 94°C for 5 sec and 72°C for 3 min, 5 cycles of 94°C for 5 sec and 70°C for 3 min, and 20 cycles of 94°C for 5 sec and 68°C for 3 min.

Paragraph 3 on page 22

Examining the sequence of the 5'-RACE-fragment indicated a double sequence, difficult to read, near the 5'-end of the fragment. However at the end of the fragment only one sequence appeared, due to a long UTR in one of the UNG-sequences but not the other. A new primer complementary to this 5'-end was designed (5'-ATGGAATTCGATTGAGATTGGCGCCTTTGG-3') (SEQ ID NO:12) and a new PCR-reaction was carried out with this, and the 5-'RACE internal primer, with the 5'-RACE fragment as template. The PCR was carried out in a final volume of 50 μl with 10 mM Tricine/KOH pH 9.2 (25°C), 15 mM KOAc, 3.5 mM $Mg(OAc)_2$, 3.75 μg/ml BSA, 0.2 mM of dATP, dCTP, dGTP and dTTP respectively, 1U Advantage cDNA Polymerase Mix (Clontech), 10ng cDNA as template and 0.2 µM upstream and downstream primers respectively. Amplification was done in a GeneAmp 9700 thermocycler (Perkin Elmer), 94°C for 1 min. followed by 30 cycles of 94°C for 30 sec, 60°C for 1 min and 72°C for 1 min.

Paragraph 3 on page 24

rcUNGΔ74:UDGL77 (5'-ACCATGGAATTCCCAAAAGCA ACGCCTGCA-3') (SEQ ID NO:13) and UDGEND2 (5'-GAGCTCGTCGACTTAGAGTGCCTCTCCAGTTTATAGG-3') (SEQ ID NO:14) and 10 ng cDNA as template.

Paragraph 4 on page 24

rcUNGΔ81:UDGL84 (5'-ACCATGGAATTCTTCGGAGAGAC TTGGAGAAGA-3') (SEQ ID NO:15) and UDGEND2 and 10 ng cDNA as template.

Paragraph 5 on page 24

rcUNGΔ74o: (5'-ATGGAATTCGCAAAAGCAACGCCTGCAGGT TTCGGAGAGACTTGGCGTCGTCAG-3') (SEQ ID NO:16) and UDGEND2 and 1 ng rcUNGΔ81o as template.

Paragraph 1 on page 25

rcUNGΔ81o: (5'-ATGGAATTCTTCGGAGAGACTTGG CGTCGTGAGCTGGCTGC-3') (SEQ ID NO:17) and UDGEND2 and 10 ng cDNA as template.

Paragraph 3 on page 29

OP5: 5'-
TCTCTCGAGAAAAGAGAGGCTGAAGCTCCCATTGACGATGA GGATGA-3 (SEQ ID NO: 18).

Paragraph 4 on page 29

NP2:
5'-GTAGAATTCGGATCCATGTCTCCTCCAGTCTAGAT-3' (SEQ ID NO:19)